On the Life-history of a new Caeoma on *Smilax aspera*, Linn.;

BY

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[illegible]

My attention has been directed for some time past to an interesting Uredine, which occurs on *Smilax aspera*, Linn., in Simla (North-West Himalayas), but I have hesitated thus long in publishing a description of it, because I hoped to have been able to complete some gaps in its life-history, and because, for reasons which will presently appear, I felt some uncertainty as to whether the uredial form was really such, or only an abnormal uredo form. My doubt on this latter point has, however, been removed since I had the opportunity lately of consulting Professor R. Hartig of Munich upon it, and of showing him my preparations, and I take this opportunity of expressing my best thanks both to him and to his assistant Freiherr von Tubeuf, not only for the [illegible] they took in this research, but also for the extreme kindness and [illegible] with which they received me during my stay in Munich. Professor Hartig quite agrees with me in regarding this hitherto somewhat doubtful form as a true Uredinum, and it now only remains for me to explain why I do not still [illegible] publishing an account of it, until I have been able to clear up some remaining doubtful points in the life-history of the parasite. I have determined to [illegible] no longer, because I cannot now foresee when I may be able to take up the investigation again, and because the facts I have already obtained appear to me to be sufficiently interesting to warrant a publication of them as they are.

The features of special interest in this Uredine are, (*a*) that it is apparently the first complete subcuticular Caeoma yet described; (*b*) the peculiar form of germination exhibited by the teleutospores; (*c*) the peculiar way in which the uredinospores are abstricted and shed; and (*d*) the suggestive resemblance exhibited by the teleutospores to the genus Gymnosporangium. But, before [illegible] these points, it is necessary first to describe the fungus generally—

General characters and natural history of the Fungus.—It will be most

convenient to begin with the aecidial stage found towards the end of June, or in July, when the heat, after the setting-in of the rains, throws out its new shoots and leaves. At this time, in certain definite localities only, although the plant is widely distributed all over the region, these newly-developed leaves, which are readily distinguishable from the older leaves of the previous year's growth by their much lighter colour, and more delicate texture, may be found attacked by an aecidium-bearing mycelium. But, although, comparatively speaking, only a very few individuals of this widely distributed plant are thus attacked, yet when a plant is attacked, it is often largely so. Only these new young leaves, and sometimes the new stems bearing them, are attacked by the aecidium, and never the older leaves or stems. When the leaf blade is attacked, conspicuous bright yellow patches are formed, more or less irregular in shape, and varying in size from a small point to about 2 cm in diameter (Fig. 1, Plate I). These patches are, moreover, considerably thickened, and bulged, with their convexities usually towards the lower surface of the leaf. The short petioles are very frequently attacked, the young shoots rarely. A single leaf may bear from one to twelve patches of invasion, and if these be carefully examined when nearly mature, minute papillæ, brownish on their summits, are seen scattered over them both on the upper and lower surfaces, and these are the aecidia, which do not open widely, as in the other hitherto described Cæomata, but only by a pore through which the aecidospores are extruded in the form of small powdery masses. A still closer examination of these patches with a lens reveals some pellucid spots, especially on the upper surface, and these are spermogonia.

Later in the season, towards the end of September, and in October, the aecidal form begins to disappear, although a few aecidial patches may still, with diligent search, be found so late as the beginning of November. The leaves which bear aecidia apparently drop off early after the aecidiospores have been shed, for in November, although a very few ripe aecidial patches may be found, no dried-up patches can be discovered on still adherent leaves. But now, in October, a new phase of the fungus is revealed. On many leaves of the same generation which bore the aecidia, but which have now become tougher and darker green, a number of uredo-pustules may be found; and it is especially noteworthy that those leaves which still, at this late date, exceptionally bore aecidia, *always* presented many of these uredo-pustules, but at some little distance from the aecidial area, with intervening healthy tissue. In other words, the uredo-pustules are formed by a distinct mycelium with, as will be later noticed, very different characters and properties. This uredo-phase of the fungus life-history is also very much more widely distributed than the aecidial, and is to be found almost everywhere, wherever the host grows, and not only in certain restricted localities as in the case of the aecidial phase. At this time when the uredo stage is beginning to show itself, the lower surfaces of the leaves attacked exhibit a few, or a very great many, slightly paled circular areas

of mycelial invasion, on each of which appear minute pus[illegible] containing yellowish brown uredospores. These invaded areas are n[illegible] in the least thickened, and, as the slight paling of the green tissues caused by the mycelium is most pronounced on the lower surfaces of the leaves, where only the pustules are formed, the affection is by no means conspicuous, and in this respect offers a very striking contrast to the aecidial affection. In the special localities where the aecidium was formerly prevalent this uredo-affection is especially prevalent, and the attacked leaves present so many foci of attack that the circular areas of each separate attack frequently merge into neighbouring areas, and thus a large area of the leaf's under-surface is uniformly paled, and bespattered irregularly with uredo-pustules. But, when the leaves are not so extensively attacked, a very peculiar and characteristic arrangement of uredo-pustules is evolved. At first, after a circular paled area has been formed by the mycelium, a uredo-pustule usually breaks out from the centre of the area. A little later a circle of other pustules is formed around the central pustule. Later still, a second larger circle, around the first circle, is formed near the margin of the paled area. But before this last circle is formed the development of teleutospores, in the form of puccinia, has commenced in the earliest formed uredo-pustules, and especially in the first circlet of these. This teleutospore formation begins to take place in November. The originally very small uredo-pustules enlarge greatly when teleutospore formation sets in, so that the neighbouring pustules of the circlet often coalesce, sometimes wholly, when a complete circular wall of teleutospores is formed round the central pustule, which does not enlarge to any great extent, and which often does not form any teleutospores, until a much later date. More usually this circular wall of teleutospores is broken in continuity at one or two places. With the onset of the winter the continued production of teleutospores goes on slowly, but apparently more or less continuously, and in the following spring the condition of the attacked leaves is as follows. Such leaves now exhibit slightly paled circular patches, varying in number from one to about twenty, without any thickening or hypertrophy. The larger patches measure from [illegible] to 7 mm. in diameter and the smallest about 2 mm. The teleutospore beds are aggregated together circularly in hemispherical dark-brown compact masses (Fig. 2, Plate I). Sometimes in the smallest paled areas there is only one such bed of spores in the centre of the discoloured area, indicating the formation in the previous autumn of only one uredo-pustule, but usually there are two or more segments of a circle, around a central space, which contains the first formed uredo-pustule now containing a few teleutospores, with remains of uredospores. When a leaf has been extensively attacked the circular arrangement, as already explained is lost, but otherwise the circular teleutospore beds are surrounded by the second circle of uredo-pustules, about nine to ten in number. This outermost circle of pustules contains frequently only old uredospores, and perhaps a few

teleutospores, though sometimes these pustules also produce many teleutospores. They do not, however, enlarge and coalesce to form another ring.

In this condition the fungus awaits a fresh season of growth for the host at the onset of the rainy season, when the young leaves are again attacked by a mycelium bearing aecidia, thus completing the circle of development.

Before proceeding now to a description of the microscopic characters of the fungus, I may mention that if a leaf bearing teleutospore beds be placed in a moist chamber for some hours, the spore beds swell very noticeably, and become lighter brown in colour, or even brownish yellow. This swelling is due, as will be more particularly explained further on, to the swelling of a gelatinous sheath, enclosing the stalks of the teleutospores, as in the case of Gymnosporangium spores. The amount of swelling, however, in the case of this Smilax fungus is much less than it is in Gymnosporangium, not so much because the gelatinous sheaths themselves swell less, but because the stalks are very considerably shorter than those of Gymnosporangium spores. This peculiar structure of the stalks of the teleutospores is interesting in its resemblance, not only to Gymnosporangium spores, but also to the teleutospores of *Puccinia Berberidis*, Montagne, occurring on the leaves of *Berberis glauca*, found on the Island Juan Fernandez, and figured by De Bary in the plate illustrating his paper on *Aecidum abietinum*[1] I will recur to these resemblances later.

Microscopic characters of the Fungus.—The mycelium which bears the uredo and teleutospores calls for no special description. It does not give rise to any hypertrophy of the host's tissues, nor does it form haustoria. The hyphæ contain colourless translucent matter, and are therefore difficult to recognise (Fig. 6, Plate I).

The aecidium-bearing mycelium has, however, a very different character. It gives rise to very marked changes in the tissues of the host (Fig 4, Plate I), with which it comes into contact, and, as the hyphæ contain orange red globules in abundance, they are readily recognisable in fresh sections. Moreover, the mycelium gives rise to the accumulation of nutritive material in the parenchyma cells among which it ramifies. The amount of hypertrophy caused in the leaf tissue is considerable, making the thickness of the lamina three times its normal size when the parasite is fully developed. The effect upon the host's tissues produced by this fungus is clearly shown in figures 4 and 5, Plate I, representing sections of the normal and attacked leaves sketched under exactly similar conditions. In the first place it will be observed that in the normal leaf there is no division of the subepidermal tissue into palisade and spongy cells, all being of the latter kind. When the leaf is attacked by the aecidial parasite, these spongy cells are greatly hypertrophied, whilst mycelial filaments permeate the tissue, sometimes destroying the cells but never apparently entering them as haustoria (Fig. 7, Plate I). In the normal leaf it is also seen that the fibrovascular bundles usually extend through the entire depth of the lamina,

[1] Botanische Zeitung, 28th November 1879, No 48, p. 846.

and these bundles are often seen to limit abruptly the extension of mycelial invasion. When the petiole is attacked it is also greatly hypertrophied, the areas of transverse sections through normal and attacked petioles being in the ratio of about 100 to 170. Such transverse sections, through attacked petioles, show that the aecidia are laid in the fundamental parenchyma tissue, below the hypoderma. The fundamental parenchyma cells are throughout considerably hypertrophied, for, whilst in transverse sections the normal cells measure in outline from 57 × 38μ to 82 × 50μ, those among mycelium measure from 95 × 63μ to 139 × 88μ.

The *teleutospores* by transmitted light are pale yellow, double-celled bodies, Puccinia, with long stalks, which, as already stated, are surrounded by a gelatinous sheath, swelling considerably in water (Fig. 13, Plate I). The free end of the upper cell is usually perceptibly thickened. The spores vary greatly in size from a total length of 74·0 to 50·8μ. The upper cell was found to vary in size from 38μ in length by 16μ in breadth to 25 × 15μ, and the lower cell from 36 × 16μ to 25 × 15μ. The spore is slightly constricted at the septum, which measures usually 14μ in breadth. These spores are detachable with difficulty, not only from their beds, but from one another, as they tend to break off in masses when scraped off by a needle. The stalks are from 2 to 2½ times as long as the whole length of the spore, *i.e.* about 125 to 156μ, and when swelled in water they are about 12μ in diameter. A nuclear space is sometimes clearly defined in each cell, but not always. The epispore is smooth and without any markings. When the stalk is swelled in water the central thin axis is clearly defined, as in Gymnosporangium. The spores germinate in water very readily, throwing out a promycelium from each cell, into which the pale yellow contents of the spore body wander, accumulating in the distal extremities (Fig. 14, Plate II), leaving the proximal parts filled with colourless fluid. The promycelium of the upper cell emerges from the apex of the cell, and that of the lower from a point near the septum, as in most Puccinia, but unlike Gymnosporangium. From this point the further developmental phenomena are curious, and until the issue of the last Botanische Zeiting,[1] which I have just received, unlike those of any hitherto described species. The observations made in this number of the Botanische Zeitung by F. Kienitz-Gerloff on the peculiar development, *at times*, of the teleutospores of *Gymnosporangium clavariæforme* add considerable interest to the Smilax fungus, for, whilst before there was only a morphological resemblance, which might well have been of little or no importance, we have now a biological resemblance, and both together suggest a closer relationship between Gymnosporangium and the other Æcidiomycetes than has hitherto been suspected. The ends of the promycelia certainly usually divide into four cells (Fig. 15*c*, Plate II) by transverse septa much in the usual way, but those cells, instead of producing sporidia, at the ends of sterigmata, round off at

[1] Botanische Zeitung, No. 25, 22nd June 1888, p. 389.

the septa, and separate, and apparently represent, sporidia. Frequently only three of the four cells formed thus separate, the fourth not maturing. These detached cells, or sporidia, have usually one or two vacuolated spaces, and measure from $14 \times 8\mu$ to $18 \times 11\mu$ (Fig. 15 *a*, *b*, *c*, Plate II). I have never, however, succeeded in getting them to germinate, unless, indeed, Fig. 15 *a*, plate II, represents the germination of a spore not yet detached; but, on the other hand, this may, and I think probably does, represent an abortive attempt at the usual sporidial formation through a sterigma. Of the numerous cultivations I examined carefully this was the only case exhibiting such growth, all the rest developing exactly as I have described and depicted. The absence of any observed germination of these sporidia makes one serious gap in the life-history of this fungus. It may also be urged that this peculiar sporidial development is due to the unsuitable media (water and a decoction of Smilax leaves) in which I placed the teleutospores. This of course is possible, but against this view I may note that it, and one other teleutospore, to which I shall presently allude, are the only ones of the many teleutospores I have observed germinating, which develope so, and that they *invariably* develope so most readily, and without any sign of hesitation. In 24 hours a cultivation presents innumerable such sporidia. The single other case to which I have alluded, is a Uromyces on *Solidago verga-aurea*, Lin., which behaves in exactly the same way, and in which, moreover, the peculiarly developed sporidia germinate readily in water. Figure 16, Plate II, represents the teleutospore before germination; Figure 17 *a*, one which has commenced to germinate, but in which the promycelium still remains undivided; and Figure 17 *b*, the cast-off terminal end of another promycelium, with four sporidia still adherent to one another, formed just as those of the Smilax fungus are, in two of which germ tubes are thrown out. I will not here encumber this paper with a detailed description of this Uromyces, but merely mention those points in support of the views I take, that this sporidial formation is a new type of normal germination.

The *uredospores* are oval, or more usually pyriform, pale yellow bodies, beset externally with very prominent spines (Fig. 12, Plate II), and so closely resemble the aecidiospores as to make it difficult to distinguish the one from the other. Figure 9, Plate II, represents some spores from leaves which were collected early in October, and preserved in botanical drying paper. The spores were sketched late in November after the leaves had been 24 hours in a moist chamber. These may be compared with fig. 8, Plate II, representing aecidiospores drawn under exactly similar conditions. Among the uredospores a few club-shaped paraphyses may always be seen. The spores are formed singly on short stalks about the length of their own long diameter (Fig. 6, Plate I). The fresh spores, after lying a few minutes in water, measure on an average $46{\cdot}5 \times 31{\cdot}7\mu$. The epispore is thickened at the broad free end. These spores do not germinate at all readily in water: only in one of many cultivations did a few germinate, throwing

out a usual, long tube, sometimes with short branches. This cultivation, made on the 11th October, contained new matured spores.

The *aecidium* is deeply placed both in the leaf and in the petiole, and is not bounded by any peridium, but by a layer of convoluted hyphæ (Fig. 4, Plate I). It opens by a narrow mouth both on the upper and lower surfaces of the leaf. The hymenium is curved and the aecidiospores are given off successively from basidia, but the ripe spores do not remain attached to one another, forming rows, as usual. On the contrary, as each spore ripens, it is cast off, and the cell below, which up to this time remained in a rudimentary condition, then grows rapidly, forming another ripe spore, and so on (Figs. 18 and 19, Plate II). The spores are pale yellow, mostly oval, with an epispore of variable thickness, thickened at one end, and beset with large coarse spines, which are deciduous. When just wetted, the spores measured on an average 43·2 × 25·6μ, varying from 36 × 28μ to 52 × 16μ. The thickness of the epispore is usually about 4μ and 6 to 10μ at the thickened end. These spores, like the uredospores, do not germinate at all readily in water. In one only of the numerous cultivations I made had a few commenced to germinate after four days. Some of these had thrown out two small germ tubes (Fig. 11, Plate II), and some only a single one (Fig. 10, Plate II). Beyond this they never went, the germ tubes were 8μ in diameter.

The *spermogonia* occur plentifully both on the upper and lower surfaces of the leaf, but more especially on the upper. They are deeply sunk below the epidermis (Fig. 3, Plate I), and through their mouths a tuft of brittle paraphyses protrude. They are of the usual structure, and measure when ripe, 145μ in depth and 157μ in breadth, the paraphyses protruding about 50μ beyond the mouths. Even in this stage, and before the inception of aecidia, the leaf tissues are greatly hypertrophied, one measurement made showing the depth of the laminal tissue in invaded areas to be 504μ against a normal depth of 296μ close by.

Concluding Remarks.—This completes the description of the fungus, and it now remains to add a few remarks on its life-history, and the affinities suggested with other Uredineæ.

I made many attempts to reproduce the aecidial form by sowing teleutospores on the young newly developed leaves at the time when the aecidial form occurs in nature, but, with one exception, these attempts failed. I do not consider the one success sufficient in itself to establish the genetic relationship, as the possibility of accidental, and unintentional, infection must, in such case, be admitted, but taken with the natural history of the fungus, the one success makes it extremely probable that we have here an autœcious uredine. I am inclined to attribute the negative results of all my other experiments to the absence of some necessary external condition, which I have not yet been able to discover, and not to the teleutosporic form being foreign to the aecidial; and I am supported in this view on two grounds, first, because I sowed the same teleutospores, during three years, on several other hosts which bore other aecidia in the neighbourhood, and, *vice versâ*, other teleutospores from other hosts

on Smilax without any result, and secondly, because even in nature, although the teleutospores are widely and plentifully distributed over the Simla region, the aecidial form is found only in certain few localities, where only, I presume, the special conditions necessary for attack exist (these localities were moist, densely wooded, and therefore very shaded). The resemblance of the teleutospores to those of Gymnosporangium induced me also to make many attempts to reproduce an Aecidium (Roestelia) on *Pyrus variolosa*, Wall., which in certain years is very common in Simla, but without success. This Roestelia is the only representative of the genus in Simla, and there can, I think, be no doubt that it is caused by the only Gymnosporangium in Simla, one on *Cupressus torulosa*, Don. I have, however, elsewhere[1] shown that, although these last two fungi must be related, I could never demonstrate it by experiment. And this case shows very forcibly how easily inoculation experiments may fail, because it is scarcely conceivable that this only Gymnosporangium, which occurs in Simla, is not related to the only Roestelia we have. There is also this parallelism between the Pyrus and Smilax Aecidia that both are relatively rare in comparison with the largely distributed and readily germinating teleutospores, and here, too, I think my inoculations have not succeeded, because some special unknown condition, comparatively rare even in nature, must be present before attack by the teleutospores can take place.

I was even more unsuccessful in attempting to produce the uredo-form by sowing with aecidiospores, for all my experiments in this direction were fruitless. The fact, however, that in nature, in October, the leaves which bore aecidia were also invariably attacked by the uredo-producing mycelium lends strong support to the view that this was the result of infection by the aecidiospores produced on the same leaf.

The total result therefore is that, although unimpeachable evidence does not exist to connect the two forms of fungus with one another, such a connection is rendered in the highest degree probable by a consideration of the facts above set forth.

In my introductory remarks I noted that I felt doubtful whether the phase I have described as aecidial were really so or not. I doubted because (1) the aecidia were unlike other aecidia in opening by a pore, (2) because the aecidiospores were not formed in rows as usual, and (3) because the aecidiospores are morphologically scarcely distinguishable from the uredospores. But, as I have said, I have now no longer any hesitation on this point, because the character and properties of the mycelium producing the aecidia are very different from those of the uredo-producing mycelium, and because the acidia are always accompanied by spermogonia, and the mycelium which produces these organs never produces teleutospores. The reason why the aecidiospores are not formed in rows may, I think, be due to the aecidia not opening widely enough to admit of this. As soon as a few aecidiospores are formed within the limited capacity of the aecidium there is no room for others until some are extruded.

[1] Journal of the Asiatic Society of Bengal, Vol. LVI, Part II, No. 3, 1887.

Lastly, a few remarks with reference to new affinities suggested by the new type of sporidial formation displayed. Had it not been for Prof. R. Hartig's discovery of the life-history of *Cæoma pinitorquum*, in connection with *Melampsora populina*, the only other Cæoma, I believe, whose life-history is known, I should naturally have been inclined to regard this new type of sporidial formation as probably peculiar to Cæoma. In the Simla region we have, so far as I know, only one Cæoma other than that on Smilax, namely, on the leaves of *Morus alba*, Lin., var. *serrata*, and it is possible that the Uromyces I have alluded to on *Solidago verga-aurea*, L., presenting the same type of sporidial formation, may be connected with it. However, whether this be so or not, Hartig's discovery shows that it is not a necessary characteristic of the Cæomata.

Kienitz-Gerloff thinks that the somewhat similarly produced spores from the promycelia of *Gymnosporangium clavariæforme* are in nature uredospores, but I do not think this view will be readily accepted. Looking at the three forms, (1) *Puccinia Berberidis*, Montague, (2) *Gymnosp. clavariæforme*, and (3) *Cæoma Smilacinis* and the Uromyces on *Solidago verga-aurea*, it appears to be more probable that we have here an evolution series, but in which direction, whether from (1) to (3) or from (3) to (1) there are not sufficient data to determine. As the process of sporidial formation is simpler in *Cæoma Smilacinis*, as I propose to call this fungus, and in the Uromyces on *Solidago verga-aurea* than in other cases, these may probably represent the earliest stages in development towards the usual mode. It is conceivable that the earliest teleutospores were hardier uredospores, which tided the fungus over unfavourable seasons, and then germinated as usual, throwing out a germ tube which directly penetrated the host (autœcious). From this might be evolved a form like *Cæoma Smilacinis*, and through this the usual form with sporidia at the ends of sterigmata. In such case *Gymnosporangium clavariæforme* would represent a form still retaining traces of a former stage of development.

The 14th July 1888.

Description of the Plates.

PLATE I.—1. *a* lower and *b* upper surface of leaf attacked by aecidial form; natural size.

2. Lower surface of leaf gathered in spring with uredo and teleutospore-pustules, showing circular arrangement; natural size.

3. Transverse section of leaf through spermogonium × 350.

4. „ through aecidium, × 150. Fig. 5 is drawn from the same section at an unattacked part.

5. Transverse section of normal leaf, × 150.

6. „ leaf through uredo-bed, × 150

PLATE II.—7. Portion of transverse section of leaf through an aecidial patch, showing mycelium between spongy cells, × 350.

8. Group of aecidiospores from dried specimen, moistened in water, × 350.

9. Group of uredospores, also from dried herbarium specimen, moistened, in outline only, × 350.

10. Aecidiospore showing commencing germination; only one germ tube, × 350.

11. Aecidiospore showing two germ tubes, × 350.

12. Fresh uredospore, in water, × 350.

13. Teleutospore in outline, showing young, still undivided promycelia from each cell, × 300.

14. Upper ends of two promycelia from same spore just before division, × 350.

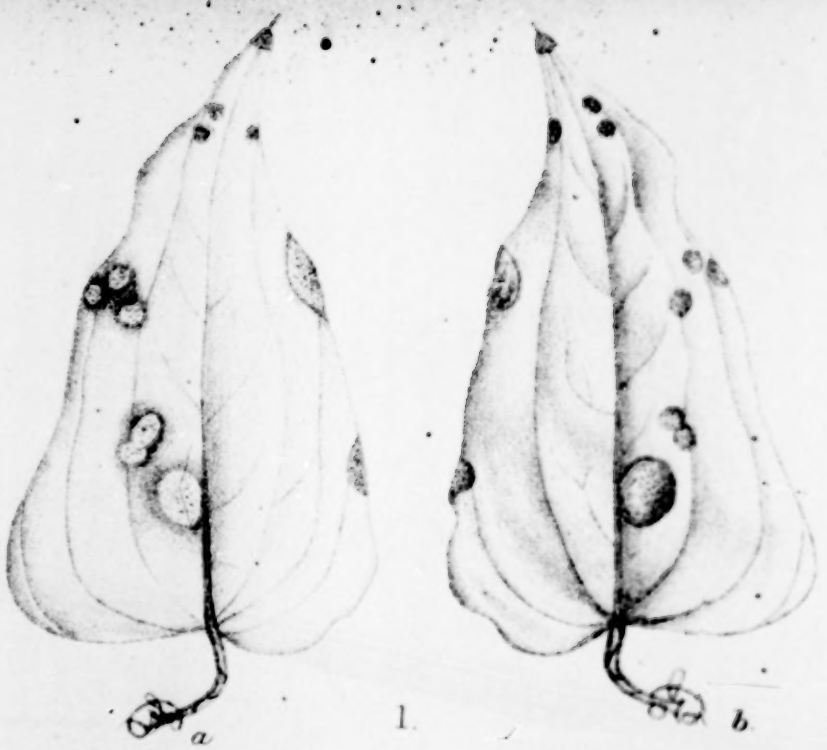
15. The same from another spore: *a*, three sporidia have been formed, one of which has thrown out a germ-tube (or abnormally a sterile sterigma?); *b*, two sporidia are ripe and ready to be cast off; *c*, one sporidium ripe and about to be cast off, while two others are still immature, × 350.

16. Uromyces (teleutospore) from leaf of *Solidago Vergaa-urea*, × 350.

17. „ ; *a*, showing promycelium still undivided; *b*, showing upper end of another promycelium consisting of four cast-off sporidia, still however attached to one another, two of which have thrown out germ tubes, × 350.

18. Section showing portion of aecidium to illustrate the manner in which aecidiospores are formed, × 350.

19. Portions from other similar sections to illustrate more clearly the same thing, × 350.



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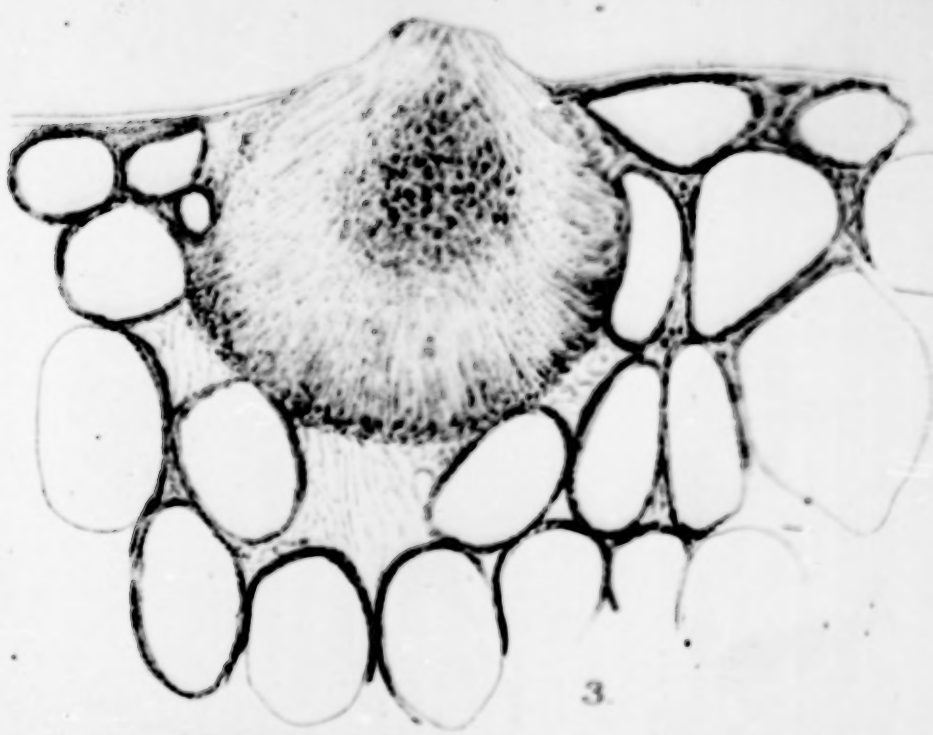

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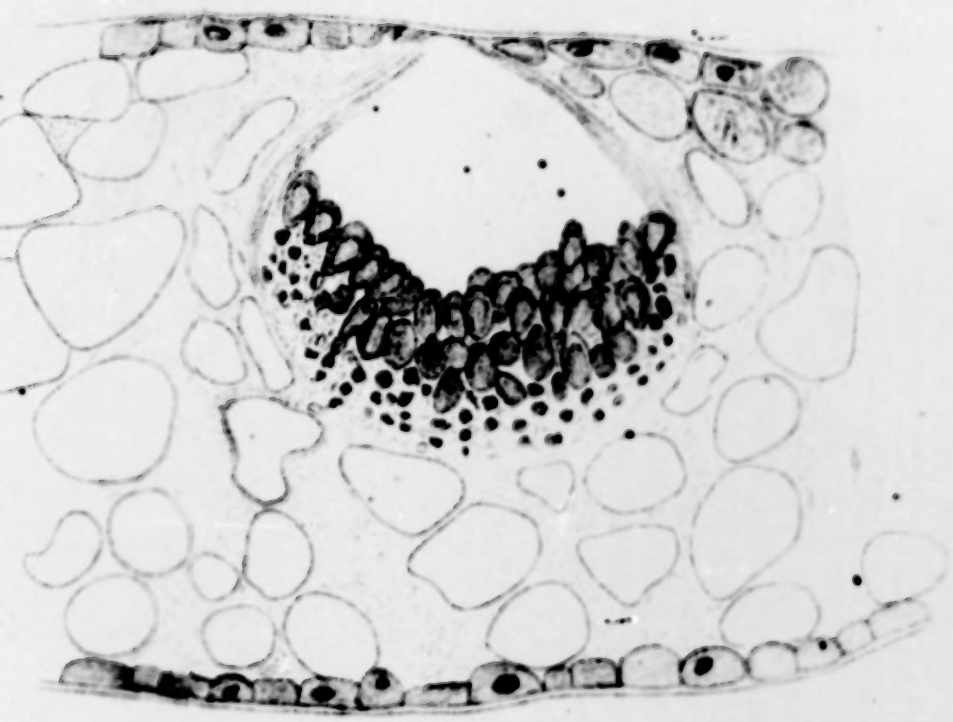

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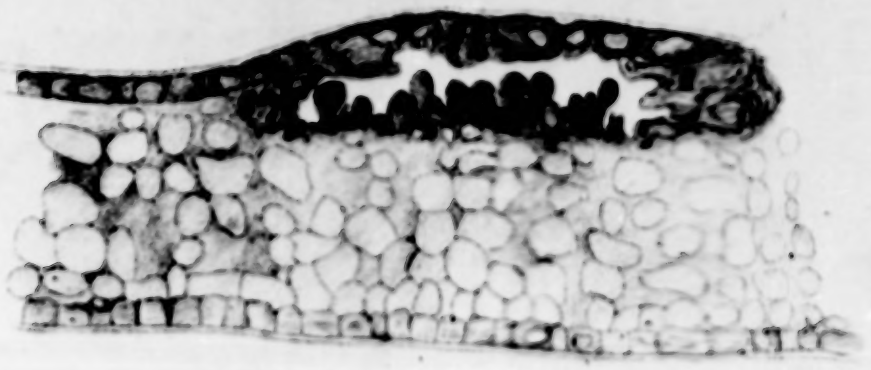
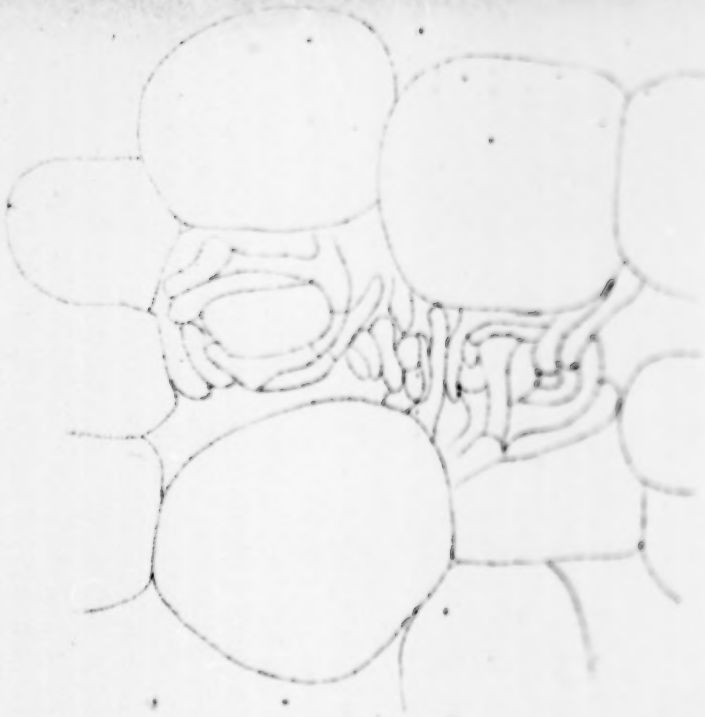


6.

A. Barclay, del.

CAEOMA SMILACINIS.

Lithographed at the Survey of India Office, Calcutta, September 1868.



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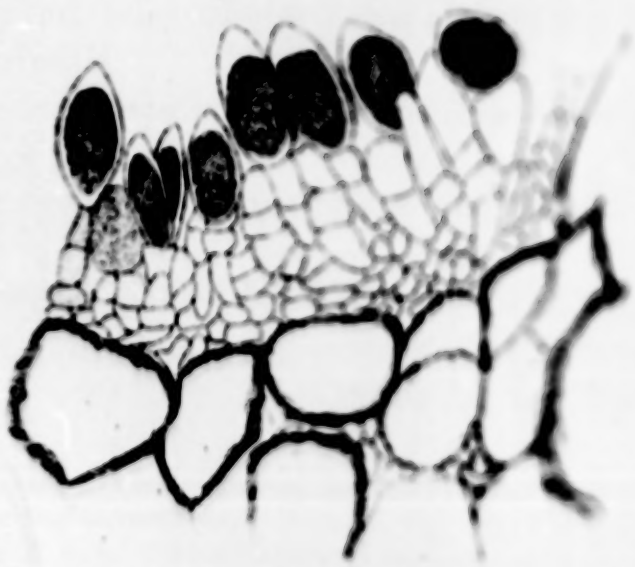

18.

A. Barclay, del.

CAEOMA SMILACINIS.

Lithographed at the Survey of [illegible]